THE RELATION OF ANTS TO THE JAPANESE BEETLE AND ITS ESTABLISHED PARASITES¹

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INTRODUCTION

It has long been known that many ants derive most of their food from dead and dying insects, and ants have been observed molesting both adult and larval stages of the Japanese beetle (Popillia japonica Newm.). Many observations and experiments have therefore been conducted in the vicinity of Moorestown, N. J., both in the field and in the insectary, to determine what relationship, if any, exists between ants (Formicidæ) and the Japanese beetle or its established parasites in the field.

Observations and diggings of the common ants, chiefly those frequenting habitats favorable to the various stages of *Popillia japonica* in this area, are recorded herein.

OBSERVATIONS OF ANT COLONIES IN 1933

In 1933 diggings were made in and near certain ant colonies in grub-infested areas in late summer and early fall to determine the grub population within the colony itself as well as in the area immediately surrounding such colonies. In general the grub population within the colonies was about the same as that near the colonies, although in some cases more grubs were found in the colony. The following detailed account of three large colonies examined in 1933 will give the reader a true picture of an average ant community in pastures of this area.

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Colony Site G-5 (Hercher's Pasture)

Hercher's pasture, located 3 miles from the Moorestown laboratory, was chosen for these studies because of its nearness to the laboratory and the willingness of the owner to permit the making of periodic surveys. Diggings in this pasture on August 8, 1933, showed the grub population to range from 1 to 32 per square foot, and according to seasonal surveys the average population for August 1933 was 9 per square foot. In the 18-square-foot area covered by this colony the average grub population was 12.2 per square foot. Grub populations in 10 diggings at random within a 25-foot radius of this infested area, but containing no ants, were as follows: 2, 8, 12, 16, 1, 8, 5, 11, 9, and 6 per square foot, or an average of 7.8 per square foot.

The species of ants in the main colony chosen was Formica pallide-fulva schaufussi var. incerta Emery, and small colonies of Lasius (Acanthomyops) claviger (Roger), and L. niger var. americanus Emery were living within the same area. The main colony of Formica covered 1 square foot. Only one entrance was visible, and the Lasius colonies were unnoticed until some soil was removed. The Formica workers had previously been seen bringing a dead larva of the eastern tent caterpillar (Malacosoma americana (F.)) to their formicary, and also a dead adult ground beetle (Calosoma sp.). They fed readily on the juices of a freshly killed Japanese beetle grub placed upon their mound, and later dragged it beneath the ground.

Colony Site G-3 (Hercher's Pasture)

This colony, of Formica fusca var. subscricea Say, was also in Hercher's pasture but 200 yards to the west and on a lower well-sodded area. During the summer the refuse heaps were examined and elytra of various Coleoptera, including one Japanese beetle, were found. Japanese beetle grubs were found at the edge of this colony throughout the summer. Diggings within the colony were limited during the summer, as the colony was under observation for feeding habits and general activities. On June 6, however, a small clump of grass within the colony boundary was removed very carefully, and one grub was found among the roots just beneath the surface. Ants were comparatively numerous

here, and when the grub was exposed they attacked it eagerly. Within one-half hour they had dragged the much weakened grub into an entrance, and ant activity again quieted down to normal.

Freshly killed grubs were placed directly upon the mound as well as very near to it. The body walls of some had previously been punctured to allow the body fluids to exude, and in such cases the ants would swarm over the remains, first feeding upon the contents and then dragging the remains into an entrance hole. The unpunctured cadavers were unmolested until foraging workers later chanced upon them and dragged them within their formicary.

When a living grub was placed upon the surface of the mound or at the edge where ants were continually passing, it was attacked by these ants and taken into their formicary. If, on the other hand, the grub was placed barely beneath the surface of the soil, it remained unmolested.

On July 14 at the colony site a living Japanese beetle was observed in combat with several ants. The beetle alighted upon the bare ground, where two ants immediately offered battle, and for half an hour a struggle was carried on during which other ants were attracted to the scene. The beetle was finally overpowered and dragged into the entrance.

On July 18 a section of the colony was excavated, revealing a Japanese beetle pupa. This pupa had been unmolested by the ants, although egg galleries and runaways formed a network around it. As soon as it was disturbed, however, ants immediately attacked and carried it away. In another section of this colony three Japanese beetle larvæ were found unharmed just below the surface. Three were also found at the edge of the main colony area. A dead Japanese beetle was also found within the colony and may have been the one observed 4 days previously.

On September 11 the remainder of this colony was examined. Systematic diggings extended 4 feet to the north and to the south of the area of concentrated activity. The colony area was shaded at times during each day by two trees growing nearby. The area at the south, being covered by shorter grass than that at the north, was more attractive to the beetles for oviposition, and consequently grub population was denser. The total area dug in this colony was 52 square feet and contained 139 grubs, or an

average of 2.6 per square foot. In the central area, where the ants were consistently most numerous, the grub population averaged 1.5 per square foot, while in the bordering bare-ground area, where fewer ants existed, only 1 grub was found per square foot, probably because bare ground is unfavorable for oviposition. If the ants had molested them, there would have been fewer grubs within the colony than in the adjacent area with identical ground cover.

Colony Site S (Haddonfield Pasture)

This colony was in a pasture near the Tavistock Golf Course, Haddonfield, N. J. It was on a sodded knoll, the surrounding area sloping toward a swamp. The ants comprising this colony were the common Lasius (A.) claviger, and were hidden completely by an old burlap bag, beneath which the ground was entirely devoid of vegetation. When examination of this colony was begun, on November 1, 1933, thousands of winged males and females were present. The area not covered by the bag was densely sodded, and because this species was more or less hypogeal, its presence was not evident. Diggings showed these ants to be established over an area of approximately 8 square feet. Grubs in this area averaged 3 to a square foot, whereas the average in a 17-square-foot area immediately surrounding was only 2.6 per square foot. Additional diggings at random around the periphery of this colony and not more than 20 feet from the colony center showed an average of 1.3 beetles per square foot. The observations made in the course of these diggings further exemplify the negative effect of this species upon grubs in the soil.

FIELD SURVEYS IN 1934 AND 1935

In 1934 surveys were continued in areas infested with ants and corresponding areas not thus infested. The data in Table 1 show that in 35 separate diggings of 1 square foot each a total of 210 grubs, or an average of 6 per square foot, was found where no ants were present, while in the same number of diggings of the same size and in the same localities with various species of ants present there was a total of 213 grubs, or 6.1 per square foot. These surveys indicate rather conclusively that at least these species of ants had no ill effect upon grubs present in the soil.

TABLE 1.—Field surveys of populations of Japanese beetle grubs in areas in New Jersey infested with ants and in surrounding uninfested areas, 1934

Date and location		Grub squar in are	e foot
of digging	•	With ants	With- out ants
September 18:		ſ 8	6
Vincentown cemetery	Formica fusca var. subsericea Say	3 4 5	7 5 9
Vincentown pasture September 19:	Pheidole pilifera (Roger) Pheidole vinelandica Forel	(4 4 3	$\begin{matrix} 0 \\ 3 \\ 2 \end{matrix}$
Vincentown pasture	Lasius niger var. neoniger Emery	$\left\{\begin{array}{cc} & 7 \\ & 3 \\ & 4 \end{array}\right.$	4 5 7
Vincentown roadside	Aphænogaster fulva subsp. aquia (Buckley)	$ \left\{ \begin{array}{c} 11 \\ 9 \\ 7 \\ 7 \\ 5 \end{array} \right. $	10 · 9 · 7 · 2 · 2
September 26: Pemberton pasture	Lasius (Acanthomyops) claviger (Roger)	$\left\{\begin{array}{c}4\\0\\2\end{array}\right.$	0 1 0
September 27:	Formica pallide-fulva subsp. schaufussi var. incerta Emery	$\left\{\begin{array}{c}1\\27\\21\end{array}\right.$	31 16
Pemberton pasture	Lasius niger var. neoniger Emery	\ 24 \ 25 \ 1	$ \begin{array}{c} 27 \\ 26 \\ 3 \end{array} $
Pemberton orchard	Formica fusca var. subscricea Say	$\begin{array}{c c} 3 \\ 4 \\ 1 \end{array}$	2 7 0
October 3:		[2	0
Haddonfield pasture	Myrmica group	1 3 0	1 0 2
October 4:		(2	4
Haddonfield pasture	Lasius (Acanthomyops) claviger (Roger)	$ \begin{cases} 0 \\ 3 \\ 5 \end{cases} $	0 2 6
Total		213	210

During June and July 1935, 42 colonies of ants were examined as well as corresponding areas containing no ants. Table 2 gives data obtained from these surveys. In both the areas containing ants and in corresponding check areas the average grub population was 4.3 per square foot. Although this was less than the population in 1934, it seems evident from these surveys that the ants had no ill effect on the grub population.

ESTABLISHED BEETLE PARASITES IN RELATION TO ANTS EFFECT OF ANTS ON ADULT TIPHIA

Ants are frequently seen foraging in the field on blossoms of wild carrot (Daucus carota L.) and on the leaves of maple trees (Acer spp.). Tiphia popilliavora Roh. obtains nectar from the former and T. vernalis Roh. feeds upon the honeydew found on the latter. Twice during the 1934 season the writer witnessed conflicts between T. popilliavora adults and the common black ant Formica fusca var. subsericea. In each case a single ant had obtained a firm hold on a female Tiphia and both fell helplessly to the ground. In one case the Tiphia gained her freedom as they struck the ground, but in the other the ant retained its hold and, after several minutes of fierce battling, with the aid of two other worker ants carried the Tiphia to the formicary. Her wings became very badly damaged before she was finally lost to sight.

In May 1934 a battle between individuals of the same species of ant and a pair of *Tiphia vernalis* adults was observed at Philmont, Pa. This male and female were mating on a maple leaf directly over a mound of these ants, when they suddenly fell to the ground, landing in the midst of ant activity, where they were instantly seized by workers. During the melee the male escaped, but the less fortunate female was injured and dragged within the formicary, where presumably she succumbed.

Since *Tiphia* adults spend considerable time in the soil and on low grass above the soil, ants probably cause the death of some. One such instance was observed at the Overbrook Golf Course in May 1934, while the author was collecting *Tiphia* for colonization. A female was just emerging from the soil, presumably to feed on honey solution sprayed on foliage nearby. Several foraging ants of *Formica fusca* var. *subscricea* were present, and a

TABLE 2.—Field surveys of populations of Japanese beetle grubs in areas in New Jersey infested with ants and in surrounding uninfested areas, 1935

Month and location of digging	Species of ant found	Grubs per square foot in areas With ants
June: Hercher's pasture	Formica pallide-fulva subsp. nitidiventris Emery	111
	Formica fusca var. subsericea Say	
Vincentown pasture	Formica fusca var. subscricea Say	$\begin{pmatrix} 9 & 14 \\ 10 & 11 \end{pmatrix}$
	Pheidole pilifera (Roger)	4 60
Marlton pasture	Lasius niger var. neoniger Emery Lasius niger var. neoniger Emery	
	Lasius (Acanthomyops) claviger (Roger)	20 00
Moorestown roadside	Tetramorium cæspitum (L.) Aphænogaster fulva subsp. aquia (Buckley)	6 9
	Formica fusca var. subsericea Say	21 00
Spring Hill Country Club,	Formica pallide-fulva subsp. nitidiventris Emerv	61
	Tetramorium cæspitum (L.) Lasius (Acanthomyops) interjectus Mayr	40,
Haddonfield pasture	Myrmica group	m 0
	Formica pallide-fulva schaufussi var. incerta	{ 4 11 5 5 9 9 9 9 9 9 9 9 9 9 9 9 9 9 9 9
	Formica fusca var. subsericea Sav	9 9

TABLE 2—(Continued)

3(+1 beetle) 3 2(+1 beet	Month and location or digging June, cont.: Medford pasture Woodbury Golf Course July: Hercher's pasture		Grubs per squ With ants $\begin{cases} 4 \\ 7 \\ 3 \\ 4 \\ 4 \\ 13 \\ 2 \\ 0 \\ 3 \\ 1 \\ 1 \\ 1 \\ 1 \\ 1 \\ 1 \\ 1 \\ 1 \\ 1$	Grubs per square foot in areas ith ants $\begin{cases} 4 & 4 & 4 \\ 7 & 5 \\ 3 & 5 \\ 4 & 5 \\ 5 & 5 \\ 2 & 2 \\ 13 & 2 & 2 \\ $
		Formica paulide-juiva schaujussi var. incerta Emery and Solenopsis molesta (Say) Lasius niger var. americanus Emery	3(+1 beetle) 3	0 $2(+1 beetle and 2 eggs)$
CC 1				000

fierce battle ensued in which the wings of the *Tiphia* became damaged, preventing her flight and enabling the ants to drag her into their nest without further opposition. Incidents of this type are exceedingly interesting to watch and may happen rather frequently, but are probably of little economic importance.

EFFECT OF ANTS ON TIPHIA COCOONS

An instance of damage to *Tiphia* cocoons in the field by *Solenopsis molesta* was observed in 1932, when several cocoons held in vials stoppered with fine wire screens and buried in the soil were riddled. These tiny yellow ants were able to make entrance into the vials through the screen. Again during the following season cocoons buried in a similar manner were destroyed.

Cocoons formed naturally in the field have also been found completely riddled with holes, and frequently the same species of ant has been associated with them. Material held in the insectary has sometimes been attacked by this ant, as will be reported under "Insectary Experiments." If colonies of *Solenopsis molesta* were more numerous in the Moorestown area, they might easily prove an important factor working against parasite establishment and subsequent increase.

EFFECT OF ANTS ON PARASITIZED GRUBS

No evidence has come to the writer's attention of ants molesting parasitized grubs in the field. It seems entirely plausible, however, that some such helpless individuals should fall prey to these industrious workers. This seems especially probable at times of advanced parasitization by certain dipterous parasites, and just prior to cocoon formation in case of hymenopterous parasites. At such times the parasitized grub lies helpless, and this as well as the parasite larva itself should be appetizing food for many ants, which often are none too particular in their choice of viands.

EFFECT OF ANTS ON FLY PUPARIA

In one colony of *Formica fusca* var. *subsericea* the remains of a *Phyllophaga* adult containing a fly puparium were found. This aroused suspicion that certain Japanese beetles containing puparia of *Centeter cinerea* Ald. might be destroyed by foraging

ants, especially since a large proportion of beetles parasitized by this fly crawl beneath dead leaves and trash to die. These places are favorite hunting grounds for many ants, and such beetles may fall victims of rapacious individuals oftener than is supposed. Observations have not materially strengthened this supposition although in 1935 one beetle bearing a Centeter egg was found in a colony of F. fusca var. subsericea. In areas of heavy parasitization by C. cinerea instances of this nature might be of significance.

INSECTARY EXPERIMENTS SPECIES OF ANTS STUDIED

Twenty-three of the most common species (or varieties) of ants, found in this area, representing 11 genera, were used in extensive insectary experiments to determine if any relationship exists between Formicidæ and *Popillia japonica*. The following species were included:

Formica pallide-fulva subsp. schaufussi var. incerta Emery Formica fusca var. subsericea Say Formica fusca var. subænescens Emery Formica sanguinea subsp. rubicunda Emery Formica truncicola subsp. integra Nylander Formica sanguinea subsp. subintegra Emery Formica neogagates Emery Formica pallide-fulva subsp. nitidiventris Emery Solenopsis molesta (Say) Lasius (Acanthomyops) claviger (Roger) Lasius (Acanthomyops) interjectus Mayr Lasius niger var. americanus Emery Lasius niger var. neoniger Emery Pheidole morrisi Forel Pheidole pilifera (Roger) Pheidole vinelandica Forel Crematogaster lineolata var. cerasi (Fitch) Tetramorium cæspitum (L.) Tapinoma sessile (Say) Monomorium minimum (Buckley) Prenolepis imparis (Say) Aphanogaster fulva subsp. aquia (Buckley)

Myrmica scabrinodis group

EXPERIMENTAL PROCEDURE

Two types of cages were used in this work and are hereafter spoken of as large cages and small cages. The former type was a wooden box 1 foot square by 2 feet deep. The upper 6 inches on two sides was of glass instead of wood, permitting observation of ant activity within the cage. The top was hinged and consisted of wire screen of very fine mesh, assuring ample ventilation and entrance of necessary moisture. All wooden parts of the cage were soaked in paraffin to prevent excessive decay when buried. In the bottom several holes 1 inch in diameter were bored and covered with wire screen similar to that used on the top, to permit the entrance and escape of moisture. Further to facilitate drainage, in some of the cages long slits were made in the sides and likewise covered with screen. However, cages without such slits remained in as good condition as the others, and in excessively wet periods were in even better condition. During such wet weather boards were placed over the cages.

These cages were buried in the soil to a depth of $1\frac{1}{2}$ feet, the glass sides remaining above ground. Grass or wheat was planted in the cages before the ants and grubs were placed there. Both crops grow well and sometimes need to be cut back to prevent undue shading of the ground. Dry leaves and a medium-sized flat rock were also placed in each cage, as many ants prefer their formicary beneath shelter, at least at the start, and hypogeic species remain almost entirely under cover.

The small type of cage consisted of two plates of common window glass placed on a vertical frame so that there was a solid bottom and a half-inch space between the plates to hold soil. A cover consisting of a wedge-shaped piece of wood retained the ants. By the use of glass sides ant activity was readily observed without disturbing the enclosed individuals. It was found advantageous to darken one side of the cage with a black cloth or heavy paper. Runways were well distributed throughout the soil, but the ants always reared their young and congregated in galleries on the darkened side.

The small cages can be kept at room temperature and ant activity maintained throughout the year. Moisture must be added every 5 or 6 weeks during the winter or in cool spring and fall

weather and at intervals of 2 to 3 weeks in the summer. Ants confined in these cages for 3 months have been removed seemingly in as vigorous condition as when first placed within. The ordinary time of confinement, however, was approximately 30 days. If ants confined in these small cages for 30 days with grubs do not molest them, it is doubtful that they would if left longer. Most species, unless very small, will tunnel throughout the soil within the first week, but some never penetrate to any great depth.

Ants and grubs were placed in each of a series of these small cages, and in some the ants were fed a candy made of white sugar and honey, whereas in others they were not fed during their confinement. The ants that were fed lived longer and were kept alive indefinitely by feeding. The absence of food did not seem to force their attentions upon the grubs but merely shortened their lives.

RESULTS

Accumulated data obtained from the numerous experiments conducted in the insectary show that 54 cages containing ants had at the time of examination at least as many grubs as the corresponding check cages without ants. Of 901 grubs placed in the cages, only 428 were accounted for after 30 days in the cages containing ants and grubs, with 36 of this number being dead. In the corresponding check cages without ants, after the same number of days and receiving the same treatment, 487 of the 901 grubs were accounted for, of which 75 were dead. In other words, 39 more dead grubs were found in cages without ants than in cages containing ants, and likewise 59 more living forms occurred in the checks than in the cages containing ants. A very slight indication of the destruction of dead remains by ants seems evident from these data, which would be of little consequence unless such remains were diseased.

In experiments in which eggs of *Popillia japonica* were placed in the soil and left for development, a total of 270 was used in experiments with ants and a corresponding number in cages without ants. A total of 102 first- and second-instar grubs was found in the cages with ants, whereas 105 grubs in the same stages were found in the cages without ants at the time of examination.

From this small difference one cannot attribute serious effects by the ants on the eggs or on the first and second instars.

In many of the cages some of the third-instar grubs became adults during the course of the experiments, thus exposing at some time the helpless prepupal and pupal stages. Ants, however, paid no more attention to these than to the more active stages.

DAMAGE TO TIPHIA COCOONS

Damage by ants to *Tiphia* cocoons being held for development has sometimes been reported. Such an instance was brought to the attention of the author in 1934, when several cocoons held in rearing trays were riddled with holes and their contents eaten by the small yellow ant *Solenopsis molesta*. These trays are divided into small compartments, and one parasitized larva is placed in each section before it is filled with soil. The trays are then stacked and held for development of the parasite larva and subsequent cocoon formation. Sometimes grubs in these trays come to the surface of the soil, and on one occasion an individual confined within a compartment was observed to be overcome by myriads of these ants. *Solenopsis molesta* is supposedly fond of fats and greases, but the writer was successful in attracting them to a sponge saturated with honey.

Owing to the slight indication noted in field studies that Tiphia cocoons are occasionally riddled by ants, as well as to similar observations in the insectary, further tests were made of such tendencies. Both the small and the large types of cages used in other experiments were tried. In each of the smaller cages, on October 3, 1934, six parasitized grubs were placed in the soil with ants, and corresponding check cages were set up without ants. Development continued, and cocoons were visible on October 25. In all cages after 3 months the ants were as active as when first placed within, and in almost every case had tunneled throughout the soil. Daily observations of several cocoons that were visible failed to indicate any damage, although the ants had runways throughout the soil and one cocoon was in their direct path for 2 months. When the soil was removed on February 15, 1935, only one cage containing individuals of Lasius (Acanthomyops) claviger gave evidence of possible damage to cocoons. In similar later experiments with this species no interference resulted.

Thirty-seven parasitized grubs were placed in each of 12 large cages and allowed to form cocoons in the presence of ant colonies. Four species of ants were used in these tests, namely, Lasius (Acanthomyops) claviger, an ant of the Myrmica scabrinodis group, Pheidole morrisi, and Tetramorium caspitum. Equal numbers of parasitized grubs were used in similar cages without ants. These cages were buried outdoors in October 1934 and left undisturbed until April 1935, at which time they were examined for cocoons. Three hundred and twenty-nine cocoons were found both in the cages containing ants and in the checks without ants, and none of these was damaged. This was 74 per cent of the total number of parasitized grubs used and indicates a negative effect of at least these species upon parasitized larvæ or the resultant cocoons.

Since Solenopsis molesta had shown a tendency to molest Tiphia cocoons, individuals were collected for insectary experiments with this species. Several cocoon cells of copper screening were placed in soil in cages containing these ants. The mesh of this screen was coarse enough to permit entrance of ants but too fine to allow soil to sift through readily. These cocoons remained in good condition and after $2\frac{1}{2}$ months only 1 out of 25 showed signs of fungus. Twenty per cent of these cocoons were riddled by this tiny ant, showing that the species might easily become a serious factor working against Tiphia establishment and subsequent increase. Fortunately, however, these ants were not generally abundant in areas containing cocoons.

Damage to Other Popillia Japonica Parasites in the Insectary

One instance of damage to insectary material other than *Tiphia* cocoons was noted in 1932, when ants of the species *Solenopsis* molesta ruined a portion of an experiment by killing flies of *Prosena sibirita* (F.) that were being held in a small cloth cage. The ants were no doubt attracted to the cage by honey which was sprayed daily upon leaves as food for the flies. After having been attracted to the scene, they played havoc with the flies as they larviposited upon the soil.

SUMMARY

To obtain information on the part played by ants (Formicidæ) in the destruction of the Japanese beetle (*Popillia japonica* Newm.) and its parasites, observations have been made of the ants occurring in the Moorestown, N. J., area, and 23 species, representing 11 genera, found to be most common have been studied in the insectary.

Ants have been observed attacking living larvæ as well as adult beetles in the field.

Comparative data from field surveys in areas infested with ants show very little difference in grub population.

Results of numerous insectary experiments testing any possible relationship existing between ants and the Japanese beetle seem to indicate little, if any, harm by the 23 species tried, either to the egg or to any subsequent stage.

Formica fusca var. subsericea Say was observed overpowering adult Tiphia in the field.

Experiments dealing with various formicid species and *Tiphia* cocoons in the soil show only one species, *Solenopsis molesta* (Say), to damage cocoons consistently. Cocoons riddled by these tiny workers have been found in the field as well as in the insectary.

A single record is given of the finding within a colony of *Formica fusca* var. *subscricea* of a beetle containing a *Centeter cinerea* egg, and of another containing a fly puparium.